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Low- and High-Resolution Gas-Chromatography-Mass Spectrometry (GC-MS) Method of Analysis for the Presence of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) in Environmental Samples

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Chapter 6

Low- and High-Resolution Gas-Chromatography–Mass Spectrometry (GC–MS) Method of Analysis for the Presence of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) in Environmental Samples

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INTRODUCTION

An environmental monitoring project is being conducted to determine if TCDD is accumulating in the environment. TCDD is an extremely toxic, chlorocrigenic, and teratogenic material (Courtney and Moore, 1971; Schwetz *et al.*, 1973; Poland *et al.*, 1971; Neubert *et al.*, 1973; Blair,) and is a possible contaminating by-product in manufacturing processes utilizing tetrachlorbenzene and in the manufacture of herbicides containing esters of 2,4,5-trichlorophenoxy acetic acid. The single oral LD_{50} of this compound in certain species of guinea pigs is six hundred parts per trillion (600 ppt) (Executive Office of President, Office of Science and Technology, 1971).

The method used for determination of TCDD in environmental samples must be extremely sensitive and specific. The high-resolution GC/MS method of analysis meets these criteria. Ultra high sensitivity

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16. ABSTRACT

An environmental monitoring project is being conducted to determine if TCDD is accumulating in the environment. TCDD is an extremely toxic, chlorocrigenic, and teratogenic material and is a possible contaminating by-product in manufacturing processes utilizing tetrachlorbenzene and in the manufacture of herbicides containing esters of 2,4,5-trichlorophenoxy acetic acid. The single oral LD_{50} of this compound in certain species of guinea pigs is six hundred parts per trillion (600 ppt).

17. KEY WORDS AND DOCUMENT ANALYSIS

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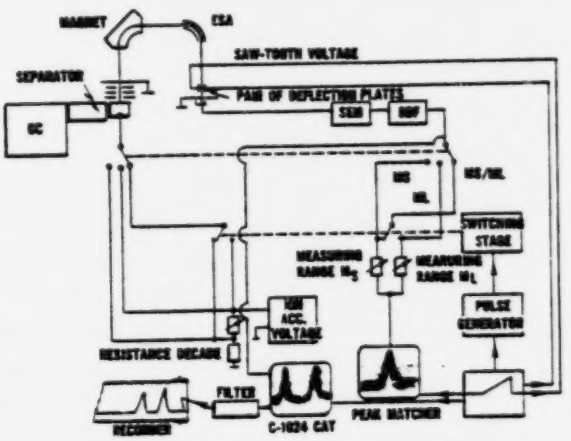

1 Block diagram of GC/MS system.

and three different degrees of specificity are provided by this technique: (1) GC retention time of TCDD, (2) double ion monitoring response, *m/e* 320–322, and (3) isotopic ratio of ^{35}Cl and ^{37}Cl. Detection limits below eight parts per trillion (8 ppt) can be obtained from 5-gm environmental samples that have been subjected to an efficient analytical cleanup procedure prior to GC/MS analysis.

The TCDD analysis procedure is based on a signal averaging technique originally developed by Biros (Biros, 1970) wherein successive scans over a narrow mass range are added together to produce an integrated mass spectrum with a greatly improved signal to noise ratio for single-ion monitoring analysis. Modifications were made to a Varian C-1024 time averaging computer (CAT) and the Varian CH-5 DF mass spectrometer to provide double-ion monitoring capabilities. A block diagram describing this system is shown in Fig. 1.

The simultaneous display and accumulation of the masses being monitored is effected as follows: By a switching pulse generated by a saw tooth voltage generator of the peak matcher unit, the ion acceleration voltage is switched between the two values, 319.8965 and 321.8936, in such manner that alternate saw tooth voltage scans are

made with the two values so that there is an alternate accumulation and display of the low mass and high mass peaks. These signals are introduced into two separate channels of the CAT. The saw tooth voltage waveform sends a trigger signal to the CAT to initiate data acquisition for each channel. The number of beam deflection scans are averaged to produce a composite, integrated, narrow-range mass spectrum which is then displayed on a strip chart recorder for mathematical analysis.

HIGH-RESOLUTION GC–MS METHOD OF ANALYSIS

A two-stage Biemann-Watson separator is used to interface a Varian model 2700 GC to the MS, thus providing an all glass inlet system. The MS vacuum system consists of a 600 liter/second mercury diffusion pump on the source and two 150 liter/second mercury diffusion pumps on the flight tube and ESA.

Optimum high resolution (9000) MS conditions are established with PFK, *m/e* 319, and the magnet current is then turned to *m/e* 328, ^{37}Cl-TCDD. One hundred picograms (100 pg) of ^{37}Cl-TCDD was injected into the GC, and the signal was monitored on the peak matcher oscilloscope to determine the specific retention time region for TCDD and the appropriate number of peak matcher sweeps necessary for data acquisition by the CAT. All pertinent data are shown in Tables I and II.

Ten percent (10%) of the 60 μl environmental sample is used for recovery analysis. Duplicate analyses are performed on the environmental sample (5%) and on a ^{37}Cl-TCDD fortified sample (5%) to determine recovery of a known amount of ^{37}Cl-TCDD that had been added before the sample was subjected to an analytical cleanup procedure. This provides the necessary recovery efficiency value for correcting the ^{35}Cl-TCDD analysis. The accumulated CAT data following each injection is displayed on a strip chart recorder for mathematical analysis.

Ninety percent (90%) of the sample extract remains for ^{35}Cl-TCDD analysis. An aliquot of the sample is fortified with a known amount of ^{35}Cl-TCDD. Duplicate analyses are performed on the environmental sample and on the fortified sample. The accumulated CAT data following each injection are displayed on a strip chart recorder for mathematical analysis. The injection sequence is sample, fortified sample, sample, fortified sample. The ^{35}Cl-TCDD residue level and limit of detection are calculated from these analyses and corrected with the recovery efficiency analysis. A low-resolution mass spectrum of TCDD is shown in Fig. 2. The main feature of the spectrum is the molecular ion cluster, *m/e* 320–322. These are the two masses scanned under high-resolution GC–MS integrating conditions to detect TCDD in ppt levels.

Table I.
GC Conditions

3' x ¼" OD x 3mm ID	
Glass column	
2% OV3 on chrom W	
Column temperature	200°c
Injection port temperature	340°c
Detector temperature	270°c
Helium flow	20 mls/min
On column injection	
Sample size 4 ul	
Concentration of TCDD standard	5 pg/ul
TCDD elution time	300 - 345 seconds
TMCS treated system at 150°c and again at 250°c as needed to remove active sites	

Table II.
MS Conditions

Source housing	Heated
Source temperature	230°c
Source pressure	5×10^{-6} - 10^{-6} Torr
Filament current	1 ma
Ion acc. voltage	3 kv
Frequency, peak monitor	maximum - 5 scans/second
Resistance decade	m/e 320 m/e 322 ratio
SDF filter	3000
ML/MB switch	0.3 V/CM
SEM voltage	3.2 KV - 10^6 to 10^7 gain
Separator temperature	270°c
Separator vonverding	250°c
Lines to GC and MS	

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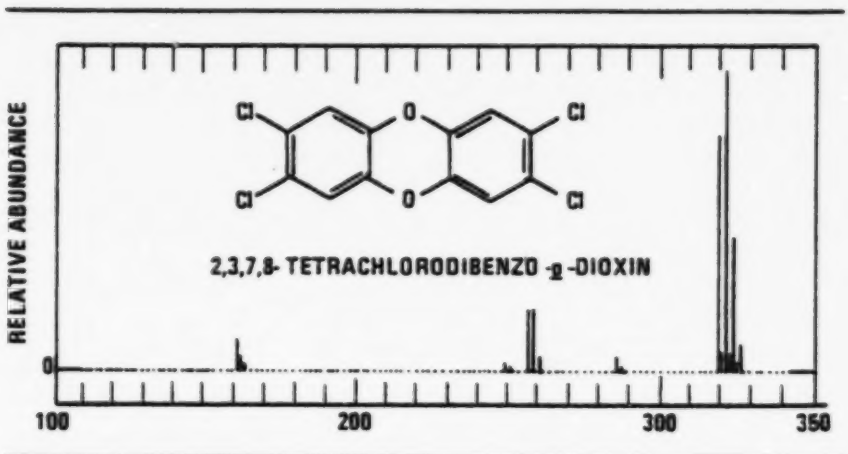

2 Low resolution mass spectrum of TCDD.

Mass spectrometric data must meet certain criteria before a sample can be reported as containing TCDD (Table 3). In addition, the TCDD peaks must be clearly defined and must have two sides with a distinct change in slope. The peaks must be clearly separated from base line and must be clearly identifiable. The TCDD content is reported as ND (not detectable) if these conditions are not met.

A tremendous amount of quality control (QC) is employed by separate laboratories, analytical cleanup and MS analysis, to provide valid analytical results. All environmental samples are fortified with ^{37}Cl-TCDD for determination of analytical cleanup efficiency. Also, environmental control samples are fortified with ^{35}Cl-TCDD, 5–100 ppt, before being subjected to the cleanup procedure. Additional QC samples containing both ^{37}Cl-TCDD and ^{35}Cl-TCDD in benzene are employed to monitor the MS analysis. These QC sample results must meet certain criteria before analysis results are reported for environmental samples.

SUMMARY

The high-resolution GC–MS method of analysis provides the necessary sensitivity, detection limit less than eight parts per trillion (8 ppt),

Table III.
Criteria Used to Define the Presence of TCDD
in Environmental Samples

1. TCDD peak must be greater than 2.5 times the rms of noise.
2. TCDD peak in both m/e 320 and m/e 322.
3. TCDD peak intensity ratio correct, 0.8 to 1.0.
4. TCDD GC retention time correct.

and specificity to monitor the environment for TCDD contamination. The use of internal standards for qualitative and quantative analysis provides high-quality results and eliminates possible sources of errors. The accuracy routinely attained for 20 to 100 pg quantitation standards and QC samples in benzene is within ± 15% of the theoretical value. An efficient analytical cleanup procedure is a prerequisite for meaningful GC–MS analysis. Quality control is very important in these analyses for picogram concentrations of TCDD. Low-resolution GC–MS screening analyses have produced some false positive results; therefore, high-resolution GC–MS must be utilized for more explicit confirmational analyses.

Our most recent GC/MS method of analysis (Harless and Oswald 1977) utilizes capillary column GC/high resolution multi-ion monitoring (*m/e* 320, *m/e* 322, and *m/e* 328) MS capabilities. The total analysis for TCDD including % recovery of ^{37}Cl-TCDD and quantification of ^{35}Cl-TCDD residue levels can be performed on two sample injections (sample and sample fortified with standard).

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DIOXIN

TOXICOLOGICAL AND CHEMICAL ASPECTS

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